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SEED PRIMING EFFECTS OF COPPER SULPHATE ON GROWTH CHARACTERISTICS OF GREEN GRAM (*Vigna radiata* L.)

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Copper sulphate is a source of copper element, a micronutrient necessary for the growth and enzymatic activities of the plant but at low concentration. To elaborate the effects of copper sulfate at different concentrations, a lab experiment was conducted according to completely randomized design with three repeats. Seed priming was done with various levels of copper sulphate (10, 20, 30, 40, 50, 60, 70 and 80 µM) along with hydro-priming and control. Seed priming with 40 µM copper sulphate gave the maximum shoot length (15.21 cm), root length (11.00 cm), fresh plant weight (9.45 g), shoot fresh weight (5.76 g), root fresh weight (3.69 g), shoot dry weight (3.13 g), root dry weight (0.76 g) and number of leaves (17.33). The results concluded that copper sulphate was useful for green gram plants but at very low concentrations and was toxic when seeds primed with high doses.
Keywords: *Leguminosae*, pulses, nitrogen fixation, copper sulphate, seed priming, green gram, stress.

INTRODUCTION

Pulses are of great importance in providing proteins to living ones. All the pulses belong to family *Leguminosae* and provide additional benefits with providing nitrogen fixing in root zone. Green gram is easily managed short duration crop as it can be raised simply in drought and saline environment. Green gram develops nodules in root and is capable of fixing nitrogen into soil (Mandal *et al.*, 2009). Despite of its high nutritional values, its cultivation and production area is not as much contrary to other leguminous crops. In Pakistan, it is conservatively grown crop but still the productivity rate is quite low due to soil and nutritional associated constraints.
To achieve good growth leading to a better yield, both macro and micronutrients are equally important (Arif *et al.*, 2006). Micronutrients have imperative role in plant growth, but these are required in less quantity. Uniform germination in crop is desired for higher yield and quality of crop (Subedi and Ma, 2005). Seed priming is one of the most conducive practice to enhance the emergence and establishment of crop stand. Seed priming is much beneficial in legumes as it substantially increases the yield of mungbean (Rashid *et al.*, 2004). Different priming techniques have been evolved which include priming with nutrients.
Copper is required by the plants for their growth but at a lower concentration, otherwise, it may become toxic (Verma *et al.*, 2011). Significant increase in overall plant growth was observed and advocated by Manivasagaperumal *et al.* (2011) when applied low concentration of Cu but growth was reduced at higher levels of applied Cu. Cu has abilities to alter plant cells and may catalyze the free radical production, for instance, oxygen reactive species which lead to oxidative burst (Hameed *et al.*, 2003). Cu is considered as pivotal element for protein component in case of several enzymes (Marschner, 1995). Cu is involved in countless physiological processes because it is an integral component of many enzymes (Hansch and Mendel, 2009). Whereas, Cu above threshold may induce chlorosis, can inhibit root growth and damages to plasma membrane (Bouazizi *et al.*, 2010).
Some of the plant species can grow in metal tainted soils (Sing *et al.*, 2008). Hajiboland *et al.* (2006) reported that plant growth, particularly, the root is most sensitive to Cu and Zn toxicity. Considering all above facts, research was conducted to evaluate effective level of Cu for seed priming which positively corelates with plant growth.

MATERIALS AND METHODS

Pot experiment was conducted with completely randomized design and three replications. NIAB Mung 2011 was used as a seed source of green gram cultivar. Each plastic pot was filled with 8 kg of soil having pH 7.7. Soil used for experiment was taken from field which was prepared for mung bean crop according to specified production technology. Nutrients added in field were N, P and K at recommended rates. Solutions of copper sulphate (10, 20, 30, 40, 50, 60, 70 and 80 µM) were made in 100 mL of distilled water. Seed priming was done for 24 hours along with hydro priming and control treatments. Seeds were soaked in clean water only for hydropriming. There was no any priming in control treatment.

After that, seeds of each treatment were dried in shade Pots were filled with fine ground soil and total 15 seeds were sown in each pot when soil was at field capacity. All the pots were labeled with their respective treatments. Sowing was done early in the morning and pots were placed under normal filed conditions, where irrigated on alternate days to avoid drying. Equal amount of water was used to irrigate each pot. After 10 days, when the plantlets were grown up, thinning was done and maintained 5 plants in each pot. Data for root length, shoot length, no.of leaves, root fresh weight, shoot fresh weight, plant weight, root dry weight and shoot dry weight was recorded twice a week. For dry weights, roots and shoots were first dried under sunlight and then in oven until a constant weight was observed for three days. Final data was taken after 30 days of thinning in which various growth parameters of the green gram were studied. Statistical analysis of Steel *et al.* (1997) was applied at 5 % probability of LSD test.

RESULTS AND DISCUSSION

***Shoot length*:** The data recorded for shoot length is demonstrated in Table 1 which predicted that the highest shoot length (15.21 cm) was measured in treatment where seeds were primed with 40 µM $CuSO_4$. However, shoot length from seed priming with 50 µM $CuSO_4$ (13.85 cm) was at par with seed priming of 40 µM $CuSO_4$. Whereas, the lowest shoot length was recorded in treatment where double amount of $CuSO_4$ (80 µM) was used for seed priming. Higher concentration of $CuSO_4$ resulted in decreased shoot length. These results are similar with Verma *et al*. (2011) as they have also reported that higher concentration of Cu resulted in decreased shoot length.

***Root length (cm)*:** Root length was also negatively affected by higher $CuSO_4$ accumulation. Data presented in Table 1 reveals that the maximum root length (11 cm) was observed when seeds were primed with 40 µM $CuSO_4$, while, the minimum root length (3.27 cm) was noted when priming was done with highest $CuSO_4$ concentration (80 µM). Reduction in root growth might be due to higher accumulation of Cu in roots as reported by Kopittke and Menzies (2006). Similarly, Souguir *et al.* (2008) also reported that highly accumulated Cu induced reduction in cell division of root.

***No. of leaves per plant*:** Leaves counted in individual plants are presented in Table 1. The data revealed that significant maximum leaves (17.33) were counted in seeds primed with 40 µM $CuSO_4$. While, the minimum leaves (7.0) were counted in treatment where seeds were primed with 80 µM $CuSO_4$. At higher concentration, leaves were less in number but were thick and small. The lowest number of leaves might be due to Cu stress which decreased the process of cell division. Thickness of leaves might be attributed to cope with the stress.

***Plant weight (g)*:** It refers to the mass of whole plant including root, shoot, leaves and pods if present. The highest plant weight (9.45 g) was recorded at 40 µM $CuSO_4$ used for priming of seeds. However, plant weight from seed priming with 50 µM $CuSO_4$ (8.54 g) was non-significant in comparison to seed priming with 40 µM $CuSO_4$. While, the lowest plant weight (1 g) was recorded where seeds were primed with 80 µM $CuSO_4$. This reduced plant weight at relatively higher $CuSO_4$ was due to less shoot height which ultimately resulted in less biomass accumulation. These results are advocated by Ali *et al.* (2002) who reported a decrease in shoot weight with toxic Cu accumulation.

***Shoot weight (g)*:** Cu helped the plants in increasing the shoot weight but up to a certain limit. Cu concentration above pertinent level started influencing negatively. The data recorded (Table 1) revealed that significantly the highest shoot weight (5.76 g) was recorded in seeds which received 40 µM $CuSO_4$ priming, while, the lowest shoot weight (0.74 g) was worked out with priming of seeds with 80 µM of $CuSO_4$. However, shoot weight from seed priming with 50 µM $CuSO_4$ (5.17 g) was non-significant when compared with seed priming of 40 µM $CuSO_4$. Similar results were reported

Table 1. Seed priming effects of copper sulphate on growth characteristics of green gram (*Vigna radiata* L.).

$CuSO_4$ (µM)	Shoot length (cm)	Root length (cm)	No. of leaves	Plant weight (g)	Shoot weight (g)	Root weight (g)	Shoot dry weight (g)	Root dry weight (g)
Control	7.65 efg	5.18 f	9.00 fg	2.00 f	1.45 fg	0.55 ef	0.53 de	0.09 g
Hydro-priming	11.54 c	8.08 c	13.66 c	5.39 c	3.37 bc	2.01 c	1.36 c	0.41 cd
10	9.04 de	6.44 de	10.66 de	3.46 de	2.37 de	1.09 de	0.73 de	0.25 ef
20	10.66 cd	7.13 d	12.00 d	4.16 cd	2.68 cd	1.48 cd	0.86 d	0.38 de
30	12.49 bc	9.24 b	15.00 bc	6.84 b	4.10 b	2.73 b	1.64 c	0.53 bc
40	15.21 a	11.00 a	17.33 a	9.45 a	5.76 a	3.69 a	3.13 a	0.76 a
50	13.85 ab	9.70 b	16.00 b	8.54 a	5.17 a	3.36 ab	2.20 b	0.64 ab
60	8.47 ef	5.57 ef	9.67 ef	2.39 ef	1.73 ef	0.65 ef	0.64 de	0.16 fg
70	6.85 fg	4.23 g	8.00 gh	1.43 f	1.01 fg	0.42 f	0.44 de	0.06 g
80	6.07 g	3.27 h	7.00 h	1.00 f	0.74 g	0.25 f	0.35 e	0.04 g
LSD	2.0559	0.8895	1.3554	1.4201	0.8475	0.6296	0.4321	0.1404

by Ali *et al.* (2002) who reported that higher Cu concentration resulted in reduce shoot weight.

Root weight (g)**:** Data presented in Table 1 illustrated that significantly the highest root weight (3.69 g) was grained by the seedlings when seeds were primed with 40 µM $CuSO_4$. However, the treatment with 50 µM $CuSO_4$ produced root weight (3.36 g) which was at par with 40 µM $CuSO_4$ priming. While, significantly the lowest root weight (0.25 g) was observed in seeds primed with $CuSO_4$ (80 µM). The reduction in root weight with increasing concentration of $CuSO_4$ is supported by Kopittke and Menzies (2006) who stated that higher accumulation of Cu reduced the root hair growth.

Shoot dry weight (g)**:** The highest shoot dry weight (3.13 g) was recorded in the treatment where seeds were primed with 40 µM Cu (Table 1). While, significantly the lowest shoot dry weight was observed in treatment where seed priming was done with 80 µM $CuSO_4$. This increase in shoot dry weight was due to higher dry matter accumulation by the seedlings whom seeds were primed with 40 µM $CuSO_4$. At higher concentration, seedling weight was reduced due to toxic effects of Cu.

Root dry weight (g)**:** Table 1 shows that the highest dry weight of root (0.76 g) was achieved by the plants where seeds were primed with 40 µM $CuSO_4$. However, dry root weight (0.64 g) from seeds primed with 50 µM $CuSO_4$ was observed at par with the seeds primed with 40 µM $CuSO_4$ concentration. The lowest value of root dry weight (0.04 g) was recorded from the treatment where seeds were primed with 80 µM $CuSO_4$.

Conclusion**:** Growth of green gram was positively promoted by priming with $CuSO_4$ as compared with control. Growth was promoted with low concentrations of $CuSO_4$ used, but seed priming with 40 µM produced the exceptional results. Priming with $CuSO_4$ above 40 µM started a decline in plant growth and reached the lowest point when seeds were primed with 80 µM $CuSO_4$. In comparison to control, the excellent results were obtained by seed priming with 40 µM $CuSO_4$, while, poor when priming was done with 80 µM $CuSO_4$.

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